CLAIMS

What is claimed is:

1. A method of purifying a fusion protein, the method comprising:

(a) contacting a sample comprising a fusion protein having a metal ion affinity peptide with a first metal ion chelate resin comprising an immobilized first metal ion;

(b) eluting any resultant bound fusion protein from said resin to produce a first eluate;

(c) contacting the first eluate with a second metal ion affinity resin comprising a second immobilized metal ion; and

(d) eluting any resultant bound fusion protein from said first and second resins to produce a product eluate comprising a purified fusion protein.

2. The method according to Claim 1, wherein the first metal ion is a hard metal ion, and the second metal ion is an intermediate metal ion.

3. The method according to claim 2, wherein the hard metal ion is chosen from Fe^{3+}, Ca^{2+} and Al^{3+}; and the intermediate metal ion is chosen from Cu^{2+}, Ni^{2+}, Zn^{2+} and Co^{2+}.

4. The method according to Claim 1, wherein the first metal ion is an intermediate metal ion, and the second metal ion is a hard metal ion.

5. The method according to Claim 4, wherein the hard metal ion is chosen from Fe^{3+}, Ca^{2+} and Al^{3+}; and the intermediate metal ion is chosen from Cu^{2+}, Ni^{2+}, Zn^{2+} and Co^{2+}.

6. The method according to claim 1, wherein the first metal ion is Co^{2+}, and the second metal ion is Fe^{3+}.

7. The method according to claim 1, wherein the first metal ion is Fe^{3+}, and the second metal ion is Co^{2+}.

8. The method according to claim 1, further comprising a wash step between steps (a) and (b) and between steps (c) and (d).

9. The method according to claim 1, wherein the contacting and eluting steps are carried out under native conditions.

10. The method according to claim 1, wherein the contacting and eluting steps are carried out under denaturing conditions.

11. A kit for purifying a protein, said kit comprising at least a first metal ion chelate resin comprising a first immobilized metal ion and a second metal ion affinity resin comprising a second immobilized metal ion.

12. The kit according to Claim 11, wherein the first metal ion is a hard metal ion, and the second metal ion is an intermediate metal ion.

13. The kit according to Claim 12, wherein the hard metal ion is chosen from Fe^{3+}, Ca^{2+} and Al^{3+}; and the intermediate metal ion is chosen from Cu^{2+}, Ni^{2+}, Zn^{2+} and Co^{2+}.

14. The kit according to Claim 11, wherein the first metal ion is an intermediate metal ion, and the second metal ion is a hard metal ion.

15. The kit according to Claim 14, wherein the hard metal ion is chosen from Fe^{3+}, Ca^{2+} and Al^{3+}; and the intermediate metal ion is chosen from Cu^{2+}, Ni^{2+}, Zn^{2+} and Co^{2+}.

16. The kit according to Claim 11, further comprising:

an extraction buffer;

a wash buffer; and

an elution buffer.

17. The kit according to Claim 11, further comprising a column.